



REPUBLIC OF NAMIBIA

MINISTRY OF AGRICULTURE, WATER AND FORESTRY

Tel: (264) 61 2087512
Fax: (264) 61 2087779

Directorate of Veterinary Services
Government Office Park
Private Bag 12022
Ausspannplatz, Windhoek

Enquiries: Dr A. Bishi
bishia@mawf.gov.na

12 July 2011

Ref: V13/3/2/7

Circular V9/2011

TO: All Deputy Chief Veterinary Officers
All Chief Veterinarians
All State Veterinarians
All Control Animal Health Technicians
All Chief Animal Health Technicians
Animal Health Technicians
All Farmers
All Auctioneers
All Food Business Operators
NNFU, NAU

RE: Brucellosis Melitensis Sampling Protocol and Maintenance of Free Status

Background

For a farm to qualify to supply sheep for export of lamb to the European Union one of the requirements is to be certified free of *Brucella melitensis* at the time of slaughter. In order to fulfill this requirement, a sample

based survey targeting **both sheep and goats** on the qualifying farm must be conducted annually following the protocol outlined below.

This Circular further outlines the procedures for the maintenance of the *Brucella melitensis* free status.

This circular replaces circulars No. V7/2005 and V13/2009.

Methodology

1. Target Holdings

All farms supplying sheep for production of lamb destined to the European Union.

2. Target Species

Both sheep and goat populations on the farm must participate in the sample survey. A proportional representation of both species must be reflected in the samples allocated to each species group.

3. Target Group

Sampling shall be targeted primarily at adult breeding males (rams and bucks) followed by breeding females. To improve the sensitivity of detecting the disease, animals that had been known to have exhibited or exhibiting clinical signs consistent with Brucellosis (e.g. epididymitis, orchitis, abortion, retained placenta, infertility etc) must be targeted for sample collection.

The other small-stock categories (non breeding) must be excluded as they are less likely to be infected.

4. Sample Size

The minimum number of serum samples to be collected at each participating holding is 35.

If the holding has less than 35 animals, then all animals on the property must be sampled.

A stratified, systematic random sampling technique should be followed to obtain the test sample. This means all breeding males should be included up to the required sample size of 35. If however, there are fewer than 35 breeding males then the difference must be made up of breeding females, selected by systematic random sampling.

An example of Systematic random sampling procedure is as follows:

- a) Establish the number of animals in the group e.g. 6 breeding males and 290 breeding females
- b) Collect blood from all 6 males, leaving an additional 29 to be taken from the breeding females
- c) To calculate the selection interval, divide 290 by 29. This gives an answer of 10 indicating that you will select every 10th animal from the group when lined up in single file.
- d) The first animal to be bled is selected randomly by choosing a number between 1 and 10. Once this number is chosen the animal corresponding with the number in the line is bled. Every tenth animal thereafter will then be sampled until the total number of blood samples has been achieved.
- e) To randomly select a number between 1 and 10, write the numbers on pieces of paper of equal size. Place the papers in a container, shake it and ask someone else to draw one paper from the container.

The sample size is designed to detect at least one positive animal for the presence of *B. melitensis* antibodies (with 95% confidence) if the sero-prevalence is 10% in the target group.

It should be noted that if less than the required number of samples are collected that particular farm will not qualify for exports of lamb to the EU until the required sample size is attained.

1. Frequency and Validity Period of Test Results

The flocks shall be tested on an annual basis ensuring that samples are collected and results obtained on or before the anniversary of the previous test result. If results are not obtained within this period, then the validity of the *B. melitensis* free status shall be suspended until the results are known.

2. Identification of Sampled Animals and Follow-up Procedure

In order to facilitate follow-up testing in case of positive or doubtful laboratory results sampled animals must be identified by suitable marking ideally using ear-tag numbers. Where this is not possible other identification methods can be applied such as paint or an indelible ink mark (number) applied on the horn.

Any follow-up investigation of doubtful laboratory results (e.g. possible false positive results) must be done in consultation with the epidemiology section and the serology section of the Central Veterinary Laboratory on a case by case basis.

3. Interpretation of Lab Results, Declaration of Freedom and Follow-up Procedure

Serum samples collected will be subjected to the Rose Bengal Test (for screening). RBT positive samples are confirmed positive or negative by the Complement Fixation Test (CFT).

A flock shall be declared free of *B. melitensis* if one or more of the tested animals test positive to the laboratory testing procedure. If one or more sheep or goats react positively, the status of the *flock* officially free from brucellosis shall be suspended and may not be recovered unless the following requirements have been fulfilled:

- (a) All sheep and goats on the farm over 6 months of age have been tested and all positive animals are removed from the farm.
- (b) Thereafter, all sheep and goats above 6 months of age on the farm have tested negative for brucellosis on two consecutive tests conducted at least three months apart.

4. Sample Collection, Labeling Handling and Shipping

Please ensure that serum is properly separated, labelled, packaged, stored and transported to the Central Veterinary Laboratory. Samples are to be accompanied to the laboratory by duly completed sampling questionnaires and herd health forms at all times. Refer to annexes 1, 2 and 5.

5. Quality Control

- (a) State Veterinarians are expected to adequately explain the contents of this circular to staff involved in sample collection so that they fully understand all the instructions as explained above at least once a year.
- (b) State Veterinarians are also expected to train staff on correct animal handling and blood sampling and handling techniques and procedures.